ESF Short Visit Grant Report
Reference Number: 4626

Purpose of the visit

Nanoparticles offer new opportunities in biomedicine and technology. Such particles are small enough to access most compartments of the body, cells and even different sub-cellular organelles. This characteristic might lead to the development of a new approach to medicine, including targeted intracellular delivery of nucleic acids, proteins or drugs. However, despite the speed at which this area is developing, relatively little is known about the ways in which these particles interact with biological systems.

Based on live cell imaging experiments, we are currently able to track individual nanoparticles and cell organelles in three dimensions. Automated detection of these objects inside cells allows us to perform an object based co-localisation analysis, and study the traffic of nanoparticles inside cells. The trajectories of the detected nanoparticles and lysosomes can be subsequently analysed to better understand the intracellular pathway of the nanoparticles, and later, the role of the protein corona in determining this. Preliminary results based on this single particle tracking platform revealed that the transfer of nanoparticles from non-acidic vesicles to lysosomes occurs via processes such as fusion between organelles and kiss-and-run events.

The aim of the short stage project was to use the high-end light microscopy facility in Prof. Jeremy Simpson's lab in UCD, Dublin, to acquire three dimensional images that will expand the knowledge about the dynamics of the intracellular traffic steps of nanoparticles. Fluorescently labeled polystyrene nanoparticles were used as model nanoparticles, and A549 cells (human lung carcinoma) were used as a model cell line.

Description of the work carried out during the visit

The experiments performed in Prof. Simpson's laboratory focused on the imaging of A549 cells (Human lung epithelium A549 cells) exposed to Polystyrene

nanoparticles. A549 cells were cultured at 37ºC in 5% CO_2 in Minimum Essential Medium (MEM, with additional L-Glutamine) supplemented with 10% Fetal Bovine Serum and 1% MEM non-essential amino acids. In order to perform the experiments, cells were seeded in live cell imaging chambers, and 24 h later were treated with LysoTracker Red dye and a short concentrated pulse of 100 nm Polystyrene nanoparticles. Imaging was performed in a spinning-disk confocal microscope equipped with a live cell imaging chamber, in order to perform experiments controlling the temperature, humidity and CO_2 levels. Three dimensional images of the cells exposed to nanoparticles were acquired at a range of incubation times after the pulse of nanoparticles. Various time samplings were used in order to explore the behaviour of the system at different time scales.

Description of the main results obtained.

The experiments performed yielded a substantial amount of three-dimensional movies of the traffic of these Polystyrene nanoparticles inside cells and the accumulation in lysosomes. The acquired data is currently being processed and will be published in a scientific article, to be submitted in the first trimester of 2012.

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